
Landoltia (Lemnaceae), a New Genus of Duckweeds

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ABSTRACT. Morphological, allozyme, and cpDNA (*rbcL*) sequence data provide evidence for the distinctness of *Spirodela punctata* from species in both *Lemna* and *Spirodela* (Lemnaceae). We propose the recognition of a new genus, *Landoltia*, to better reflect current phylogenetic concepts in the Lemnaceae. *Landoltia* is distinguished by its reduced frond prophyllum, frond nerves (3 to 7), roots (up to 7), root tracheids, external anther locules, and also by well-supported molecular evidence provided by allozymes and cpDNA sequences. The new combination *Landoltia punctata* is made to accommodate this taxonomic modification.

Duckweed classification remains equivocal because phylogenetic relationships are difficult to reconcile in this diminutive, reduced family (Les et al., 1997b). The most comprehensive phylogenetic hypotheses for duckweed taxa were presented in a monograph by Landolt (1986) and form the basis of modern classification for the family Lemnaceae.

In the most recent monograph of the duckweed family (Lemnaceae), Landolt (1986) recognized four genera: *Lemna*, *Spirodela*, *Wolffia*, and *Wolffiella*. Few additional genera have been proposed in past taxonomic treatments of duckweeds. One example is *Staurogeton* Reichenbach, which was elevated from subgeneric to generic rank by Schur (1866) to accommodate the morphologically distinctive *Lemna trisulca* L. (Landolt, 1986). Most contemporary classifications continue to assign this taxon to the genus *Lemna*. Den Hartog and van der Plas (1970) subdivided *Wolffiella* to create the two genera *Pseudowolffia* and *Wolffiopsis*. Few taxonomic treatments recognize either of these segregate genera as distinct from *Wolffiella*. To our knowledge, division of either *Spirodela* or *Wolffia* into subsidiary genera has not yet been suggested, although some authors have transferred certain species from these genera into either *Lemna* or *Wolffiella*. Landolt's recent generic concept of *Spirodela* recognizes a paraphyletic taxon, with *S. polyrhiza* and *S. intermedia* as sis-

ter species, but with *S. punctata* associating with *Lemna* (Fig. 1A).

Although Landolt's evolutionary trees reflect a keen understanding of duckweeds and a comprehensive evaluation of published taxonomic literature on the group, they were constructed using non-explicit phylogenetic methods, i.e., not by cladistic analyses. We have re-analyzed results of these earlier studies using cladistic methodologies to test hypothetical relationships proposed by Landolt. In addition, we have generated several molecular data sets to supplement the clearly limited number of phylogenetically informative characters available for these morphologically simple plants. Although this work remains in progress, the results of our preliminary analyses of morphological, biochemical, allozyme, and DNA (*rbcL*) sequence data (Crawford & Landolt, 1993, 1995; Crawford et al., 1995, 1997; Crawford et al., 1996; Les et al., 1994, 1997a, 1997b) are reasonably concordant with Landolt's classification, but differ in a number of details from his phylogenetic diagrams. In particular, generic subdivisions used in his classification of Lemnaceae are inconsistent with results of these phylogenetic analyses and merit reconsideration. Herein we summarize evidence that warrants the taxonomic segregation of a new duckweed genus. We consider this taxonomic action essential for a classification that reasonably depicts our best, current estimate of phylogenetic relationships in the Lemnaceae.

A specific diagram of intergeneric duckweed relationships that summarizes the phylogenetic trees originally appearing in Landolt (1986) was provided to us by E. Landolt. We compared these hypothetical relationships to published allozyme studies (Crawford & Landolt, 1993) and to previous cladistic analyses of morphological, anatomical, and biochemical data (Les et al., 1997b). Intergeneric duckweed relationships were also examined using preliminary results of a phylogenetic analysis of

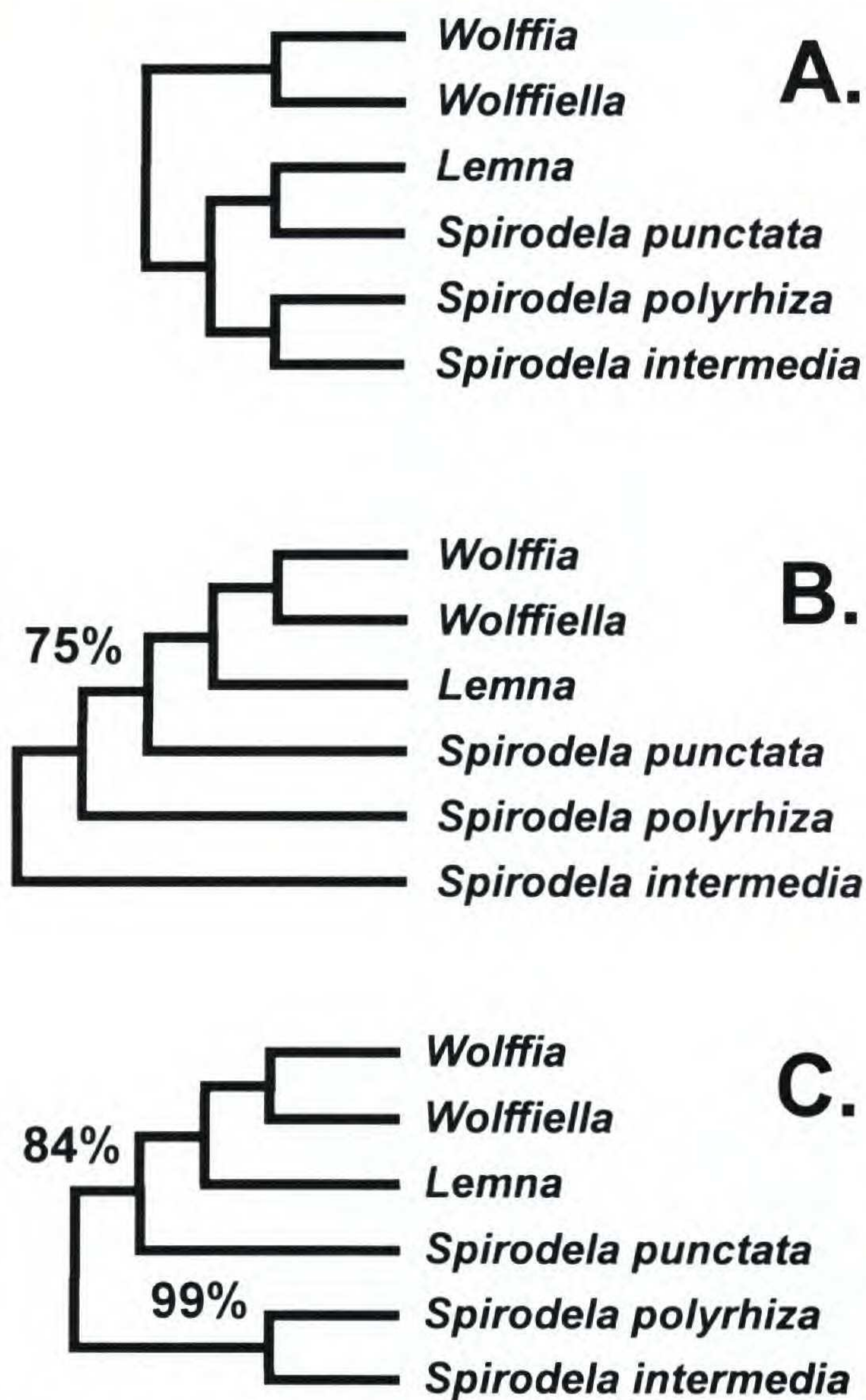


Figure 1. Phylogenetic position of *Spirodela punctata* as indicated in several recent studies. —A. Landolt (1986) hypothesized the association of *S. punctata* with *Lemna*, in a paraphyletic concept of *Spirodela*.—B. Non-molecular data (Les et al., 1997b) place *S. punctata* in a separate clade from *Lemna*, but also apart from other *Spirodela* species (bootstrap % shown). —C. Molecular (*rbcL*) data provide strong support (bootstrap % shown) for the distinctness of *S. punctata* from either *Lemna* or *Spirodela* (Les et al., 1997b). All evidence points to an isolated position of *S. punctata* in the Lemnaceae, and its recognition as a distinct genus is compatible systematically with any of these results.

rbcL sequence data for the Lemnaceae (Les et al., 1997b).

Non-molecular data (Fig. 1B) resolve the entire genus *Spirodela* as paraphyletic. However, *S. punctata* lies distinct from the other *Spirodela* species as a separate branch, and with good internal support (75% bootstrap value). Strong bootstrap values (99% and 84%, respectively) from molecular (*rbcL*) data (Fig. 1C) support *S. polyrhiza* and *S. intermedia* as sister species, and also their distinction from *S. punctata*. Allozyme data (Crawford & Landolt, 1993) show a moderate genetic identity between *S. polyrhiza* and *S. intermedia* ($I = 0.404$), yet they share no electrophoretically detectable alleles with *S. punctata* ($I = 0.000$). Various morphological features (Table 1) are consistent with a phylogenetic position for *S. punctata* intermediate between *Lemna* and other *Spirodela* species. By inspection of these features, the species *S. punctata* is not only morphologically distinct from both *Lemna* and *Spirodela*, but intermediate and transitional between these genera.

Presently, *Spirodela* Schleiden comprises three distinct species: *Spirodela intermedia* W. Koch, *S. polyrhiza* (L.) Schleiden, and *S. punctata* (G. Meyer) C. H. Thompson. Landolt (1986) placed the former two species into *Spirodela* sect. *Spirodela*, and segregated the latter in section *Oligorrhizae* W. Koch. *Spirodela punctata* has been taxonomically problematic because it possesses features similar to both *Spirodela* and *Lemna* (Table 1; Landolt, 1986).

Meyer (1818) originally named *Spirodela punctata* as *Lemna punctata*, but it was not until 50 years later that Hegelmaier (1868) transferred the taxon (as *S. oligorrhiza*) to *Spirodela*. In Meyer's time, all Lemnaceae were included in the genus *Lemna*. The new genera *Spirodela* and *Wolffia* were created in 1839 and 1844, respectively, and *Wolffiella* was established in 1895 (Landolt, 1986). It is

Table 1. Morphological features compared among species of *Spirodela* and *Lemna*. *Spirodela punctata* is intermediate between *Lemna* and other *Spirodela* species for the character states indicated (from Landolt, 1986, 1998; Shih, 1979).

Feature	<i>S. intermedia</i> ; <i>S. polyrhiza</i>	<i>S. punctata</i>	<i>Lemna</i>
Prophyllum at base of frond	present	present, but reduced	absent
No. of veins in frond	7 to 16	3 to 7	1 to 5
No. of roots	7 to 21	1 to 7 (12)	1
Root tracheids	extend to tip	basal only	absent
Dorsal meristem of new fronds	on one side ¹	on both sides	on both sides
External anther locules	do not extend above internal locules	extend slightly above internal locules	extend above internal locules

¹ Lateral on other side.

the merit of Hegelmaier (1868, 1895) to have clearly separated the genera *Lemna* and *Spirodela*. He also demonstrated accurately and comprehensively the special position of *S. punctata* (called *S. oligorrhiza*) within the genus *Spirodela*.

Spirodela punctata is very polymorphic in relation to size, pigmentation, number of roots, and veins. The high level of variability led Hegelmaier (1895) to distinguish two species (*S. oligorrhiza* and *S. pusilla*) and to describe three other species of questionable status. Growth experiments (Landolt, 1986; Landolt & Kandeler, 1987) and allozyme studies (Crawford & Landolt, 1993) have demonstrated that genetic variation in *S. punctata* is rather limited, and many of the differences used to distinguish former taxa are induced environmentally.

Thompson transferred *L. punctata* to *Spirodela* in 1898. Landolt (1986) observed that some authors have merged the genera *Lemna* and *Spirodela* because of their similar appearance and because the features of *S. punctata* are transitional between the genera. Yet, he clearly differentiated *Spirodela* (including *S. punctata*) from *Lemna* by the reduced prophyllum at the base of its fronds, druse crystals, pigment cells, multiple roots, better developed tracheids, and other anatomical/morphological features (Landolt, 1986). To our knowledge, the generic distinctness of *S. punctata* from both *Spirodela* and *Lemna* has not been suggested previously.

We have now examined relationships of duckweed genera using morphological, anatomical, flavonoid, allozyme, and *rbcL* sequence data. As summarized in Figure 1, analyses of these data sets indicate that *Spirodela punctata* represents an isolated clade distinct from both *Spirodela* and *Lemna*. Cladograms constructed from either morphological or flavonoid data (or their combination) show high internal support (75–97% bootstrap values) for the distinctness of *S. punctata* from section *Spirodela* (Les et al., 1997b) and support Landolt's original phylogenetic concept that recognized *Spirodela* as paraphyletic with respect to the position of *S. punctata* (Landolt, 1986).

Allozyme data (Crawford & Landolt, 1993, & unpublished) indicate a complete lack of genetic identity between *S. punctata* and any species in either *Lemna* or *Spirodela*, yet the two species of *Spirodela* sect. *Spirodela* do retain a moderate genetic identity. Chloroplast DNA (*rbcL*) sequence data (Les et al., 1997b) resolve *S. punctata* in a clade between *Spirodela* and *Lemna* but not within either genus. In summary, these results echo the transitional nature and unsettled taxonomic status of *S. punctata* manifest in prior systematic inves-

tigations. Phylogenetically, our broad-based/wide-ranging studies indicate that *S. punctata* is indeed transitional between, but not a member of either *Lemna* or *Spirodela*. It is for this reason that we establish a new genus to better reflect this revised hypothesis of duckweed relationships.

Landoltia D. H. Les & D. J. Crawford, gen. nov.
TYPE: *Lemna punctata* G. Meyer: Prim. Fl. Esseq. 262. 1818. ≡ *Landoltia punctata* (G. Meyer) D. H. Les & D. J. Crawford.

Herbae ex radicibus 2 ad 7 (raro 1 vel 8 ad 12) usque ad 7 cm longis, omnibus prophyllum perforantibus; turionibus absentibus. Frondes in summa aqua natantes, ovatae ad lanceolatae, 1.5–2.0-plo longiores quam latiores, supra nitidae viridesque serie mediana papillarum ornatatae, subtus laeves rubraeque; nervis 3 ad 7. Flores infrequentes; antherae loculis externis super internos positae. Fructus ala laterali in parte supera praedita; seminibus 1 vel 2 manifeste 10 ad 15-costatis.

Roots 2 to 7 (rarely 1 or 8 to 12), up to 7 cm long, all perforating the prophyllum. Turions absent. Fronds floating on the surface of the water, ovate to lanceolate, 1.5–2 times longer than wide, above shining and green with a medial series of papillae, below smooth and red; veins 3 to 7. Flowers infrequent; external locules of the anther above the internal locules. Upper part of fruit with a lateral wing; seeds 1 or 2 with 10 to 15 distinct ribs.

Landoltia punctata (G. Meyer) D. H. Les & D. J. Crawford, comb. nov. Basionym: *Lemna punctata* G. Meyer, Prim. Fl. Esseq. 262. 1818. *Spirodela punctata* (G. Meyer) C. H. Thompson, Rep. (Annual) Missouri Bot. Gard. 9: 28. 1898. TYPE: Chile. Tierra del Fuego Island, Orange Harbor, leg. Wilkes expedition 1838 (neotype, US not seen; isoneotypes, DS, GH, KANU, MO not seen).

Lemna oligorrhiza Kurz, J. Linn. Soc., Bot. 9: 267. 1866. *Spirodela oligorrhiza* (Kurz) Hegelmaier, Die Lemnaceen 147. 1868. TYPE: India. Calcutta, Kurz 1865 (holotype, CAL? not seen; isotypes, K, MEL, U not seen).

The generic name *Landoltia* commemorates Elias Landolt for his outstanding contributions to the systematics and biology of Lemnaceae in his more than 45 years of research on duckweeds.

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